

IN THE CLAIMS

This listing of claims replaces all prior versions, and listings, in this application.

1. (currently amended) A method for determining the amount of template nucleic acid present in a sample comprising:

- i) bringing into association with the sample all the components necessary for nucleic acid amplification, and all the components necessary for a bioluminescence assay for nucleic acid amplification including:
 - a) a nucleic acid polymerase,
 - b) the substrates for the nucleic acid polymerase,
 - c) at least two primers,
 - d) a thermostable luciferase,
 - e) luciferin,
 - f) ~~optionally~~ ATP sulphurylase, and
 - g) ~~optionally~~ adenosine 5' phosphosulphate;[[.]]

and subsequently:

- ii) performing a nucleic acid amplification reaction of the target nucleic acid involving more than one cycle of amplification;
- iii) monitoring the intensity of light output from the bioluminescence assay; ~~reaction~~, and
- iv) determining the amount of template nucleic acid present in the sample.

2. (previously presented) A method according to claim 1, wherein at least ii) and iii) are carried out in a sealed vessel.

3. (previously presented) A method according to claim 1, wherein in iii) the intensity of light output is monitored during the nucleic acid amplification reaction.

4. (previously presented) A method according to claim 1, wherein iii) further includes producing a data set of intensity of light output as a function of time.

5. (previously presented) A method according to claim 4, wherein the amount of template nucleic acid present is determined by measuring from the data set the time taken to reach a point at which the rate of change of intensity of light output changes significantly.

6. (currently amended) A method according to claim 1, ~~wherein for determining the~~ amount of template nucleic acid present in the sample is determined before at the ~~beginning of the nucleic acid amplification reaction of ii).~~

7. (currently amended) A method according to claim 1, ~~wherein for determining the~~ amount of template nucleic acid present in the sample is determined after as a result of the nucleic acid amplification reaction of ii).

8. (previously presented) A method according to claim 5, wherein the amount of template nucleic acid present is determined by measuring from the data set the time taken to reach a point at which the intensity of light output begins to increase.

9. (previously presented) A method according to claim 5, wherein the amount of template nucleic acid present is determined by measuring from the data set the time taken to reach a point at which the intensity of light output is at a maximum.

10. (previously presented) A method according to claim 5, wherein the amount of template nucleic acid present is determined by measuring from the data set the time taken to reach a point at which the rate of decrease of intensity of light output increases.

11. (previously presented) A method according to claim 5, wherein the amount of template nucleic acid present is determined by measuring from the data set the time taken to reach a point at which the rate of decrease of intensity of light output decreases.

12. (previously presented) A method according to claim 5, wherein the amount of template nucleic acid present is determined by measuring from the data set the time taken to reach a point at which the intensity of light output reaches or crosses a predetermined level.

13. (previously presented) A method according to claim 8, wherein the thermostable luciferase that is brought into association with the sample in i) is a reversibly-inhibited luciferase.

14. (previously presented) A method according to claim 1, wherein iv) further comprises comparing the intensity of light output to the intensity of light output from a control in which the sample comprises a known amount of template nucleic acid.

15. (currently amended) A method according to claim 1 for determining whether the template nucleic acid is present in the sample, wherein whether the template nucleic acid is present in the sample is determined by measuring from the data set whether the intensity of light output reaches or crosses a predetermined level.

Claim 16 (canceled)

17. (original) A method according to claim 15, wherein an increase in the intensity of light output relative to the predetermined level indicates the presence of template nucleic acid in the sample.

18. (original) A method according to claim 15, wherein a decrease in the intensity of light output relative to the predetermined level indicates the presence of template nucleic acid in the sample.

19. (currently amended) A method according to claim 15 [[16]], wherein whether the template nucleic acid is present in the sample is determined by measuring from the data set whether the intensity of light output reaches or crosses the predetermined level within a predetermined length of time following the start of the amplification reaction of ii).

20. (previously presented) A method according to claim 1, wherein iv) further comprises comparing the intensity of light output to the intensity of light output from a control in which no amplification has taken place.

21. (original) A method according to claim 20, wherein a decrease in the intensity of light output relative to a control reaction in which no amplification has taken place indicates the presence of template nucleic acid in the sample.

22. (previously presented) A method according to claim 1, wherein the nucleic acid amplification reaction of ii) is a low temperature thermocycling amplification method in which the cycling temperature range does not exceed 75°C.

23. (previously presented) A method according to claim 1, wherein the nucleic acid amplification reaction of ii) is carried out isothermally.

24. (previously presented) A method according to claim 23, wherein the nucleic acid amplification reaction of ii) is carried out within a temperature range that does not exceed 75°C.

25. (previously presented) A method according to claim 23, wherein the nucleic acid amplification reaction of ii) is carried out at a constant temperature at which the components of the amplification reaction and the bioluminescence assay are stable.

26. (previously presented) A method according to claim 23, wherein the nucleic acid amplification reaction of ii) is carried out at more than one temperature within the temperature range in which the components of the amplification reaction and the bioluminescence assay are stable.

27. (previously presented) A method according to claim 26, wherein the nucleic acid amplification reaction of ii) is started at a higher temperature and subsequently dropped to a lower temperature.

28. (currently amended) A method according to claim 1 adapted for use in medical diagnostics.

29. (currently amended) A method according to claim 1 adapted for use in determining whether a pathogen is present in a sample.

30. (currently amended) A method according to claim 1 adapted for determining whether a particular nucleic acid sequence is present in an organism's genetic code.

31. (currently amended) A method according to claim 30 adapted for determining whether the nucleic acid to which the template nucleic acid originates has been genetically modified.

32. (currently amended) A method according to claim 1 adapted for determining whether an organism is present in a sample.

33. (currently amended) A method according to claim 1 adapted for use in immuno-nucleic acid amplification technology.

Claims 34-37 (canceled)

38. (new) A method for determining the amount of template nucleic acid present in a sample comprising:

- i) bringing into association with the sample all the components necessary for nucleic acid amplification, and all the components necessary for a bioluminescence assay for nucleic acid amplification including:
 - a) a nucleic acid polymerase,
 - b) the substrates for the nucleic acid polymerase,
 - c) at least two primers,
 - d) a thermostable luciferase, and
 - e) luciferin;

and subsequently:

- ii) performing a nucleic acid amplification reaction of the target nucleic acid involving more than one cycle of amplification;
- iii) monitoring the intensity of light output from the bioluminescence assay; and
- iv) determining the amount of template nucleic acid present in the sample.